```
      (FILE 'HOME' ENTERED AT 06:49:49 ON 29 MAR 2004)

      FILE 'MEDLINE, EMBASE, BIOSIS, CAPLUS' ENTERED AT 06:50:47 ON 29 MAR 2004
                  E COLLIEC SYLVIA/IN,AU
L1             10 S E1-3
                  E JOUAULT SYLVIA/IN,AU
L2              3 S E1-4
L3             13 S L1 OR L2
                  E FISCHER ANNE/IN,AU
L4             79 S E4-10
                  E DURAND PATRICK/IN,AU
L5             90 S E2-4
                  E JOZEFONVICZ JACQUELINE/IN,AU
L6            120 S E1-5
                  E LETOURNEUR DIDIER/IN,AU
L7            214 S E1-5
L8             30 S MILLET JEAN/IN,AU
                  E MILLET JEAN/IN,AU
L9             70 S E2-16
L10           524 S L3 OR L4 OR L5 OR L6 OR L7 OR L9
L11        174430 S POLYSACCHARIDE
L12          3938 S FUCUS
L13           562 S FUCAN
                  SET PLURALS ON
L14        174430 S POLYSACCHARIDE
L15        148221 S ANTICOAGULANT
L16        235588 S THROMBOSIS
L17         84753 S ANTITHROMB?
L18        565869 S L11 OR L12 OR L13 OR L15 OR L16 OR L17
L19           224 S L18 AND L10
L20           111 S L19 AND L15
L21            60 S L20 AND L17
L22        959597 S SULPHAT? OR SULFAT?
L23            36 S L21 AND L22

=>
```

L23 ANSWER 1 OF 36 MEDLINE on STN
ACCESSION NUMBER: 2003169606 MEDLINE
DOCUMENT NUMBER: PubMed ID: 12649349
TITLE: Low-molecular-weight fucoidan promotes therapeutic revascularization in a rat model of critical hindlimb ischemia.
AUTHOR: Luyt Charles-Edouard; Meddahi-Pelle Anne; Ho-Tin-Noe Benoit; Colliec-Jouault Sylvia; Guezennec Jean; Louedec Liliane; Prats Herve; Jacob Marie-Paule; Osborne-Pellegrin Mary; **Letourneur Didier**; Michel Jean-Baptiste
CORPORATE SOURCE: Institut National de la Sante et de la Recherche Medicale U460, CHU X. Bichat, Paris, France.
SOURCE: Journal of pharmacology and experimental therapeutics, (2003 Apr) 305 (1) 24-30.
Journal code: 0376362. ISSN: 0022-3565.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200304
ENTRY DATE: Entered STN: 20030416
Last Updated on STN: 20030423
Entered Medline: 20030422

AB The therapeutic potential of low-molecular-weight (LMW) fucoidan, a **sulfated polysaccharide** extracted from brown seaweed devoid of direct **antithrombin** effect, was investigated in vitro and in a model of critical hindlimb ischemia in rat. In vitro results showed that LMW fucoidan enhanced fibroblast growth factor (FGF)-2-induced [(3)H]thymidine incorporation in cultured rat smooth muscle cells. Intravenous injection in rats of LMW fucoidan significantly increased the stromal-derived factor (SDF)-1 level from 1.2 +/- 0.1 to 6.5 +/- 0.35 ng/ml in plasma. The therapeutic effect of LMW fucoidan (5 mg/kg/day), FGF-2 (1 micro g/kg/day), and LMW fucoidan combined with FGF-2 was assessed 14 days after induction of ischemia by 1) clinical evaluation of claudication, 2) tissue blood flow analysis, 3) histoenzymology of muscle metabolic activity, and 4) quantification of capillary density. Both LMW fucoidan and FGF-2 similarly improved residual muscle blood flow (62.5 +/- 6.5 and 64.5 +/- 4.5%, respectively) compared with the control group (42 +/- 3.5%, p < 0.0001). The combination of FGF-2 and LMW fucoidan showed further significant improvement in tissue blood flow (90.5 +/- 3%, p < 0.0001). These results were confirmed by phosphorylase activity, showing muscle regeneration in rats treated with the combination of FGF-2 and LMW fucoidan. Capillary density count increased from 9.6 +/- 0.7 capillaries/muscle section in untreated ischemic controls to 14.3 +/- 0.9 with LMW fucoidan, 14.5 +/- 0.9 with FGF-2, and 19.1 +/- 0.9 in combination (p < 0.001). Thus, LMW fucoidan potentiates FGF-2 activity, mobilizes SDF-1, and facilitates angiogenesis in a rat model. This natural compound could be of interest as an alternative for conventional treatment in critical ischemia.

L23 ANSWER 2 OF 36 MEDLINE on STN
ACCESSION NUMBER: 2002487176 MEDLINE
DOCUMENT NUMBER: PubMed ID: 12297128
TITLE: Effect of fucoidan on fibroblast growth factor-2-induced angiogenesis in vitro.
AUTHOR: Matou Sabine; Helley Dominique; Chabut Delphine; Bros Andree; **Fischer Anne-Marie**
CORPORATE SOURCE: INSERM U428, Universite Paris V, Hopital Europeen Georges Pompidou, 20 rue Leblanc, 75908 Paris Cedex 15, France.
SOURCE: Thrombosis research, (2002 May 15) 106 (4-5) 213-21.
Journal code: 0326377. ISSN: 0049-3848.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200308
ENTRY DATE: Entered STN: 20020926
Last Updated on STN: 20030823
Entered Medline: 20030822

AB Fucoidans are **sulfated polysaccharides** extracted from brown marine algae. A purified fucoidan fraction exhibits the same venous **antithrombotic** activity as heparin in rabbits, but with a lower **anticoagulant** effect. Because of its heparin-like structure, we postulated that fucoidan might modulate heparin-binding angiogenic growth factor activity. We thus studied its effect, at **antithrombotic** concentrations, on fibroblast growth factor (FGF)-2-induced proliferation

and differentiation of human umbilical vein endothelial cells. The fucoidan effect on endothelial cell differentiation was evaluated by studying the expression of surface proteins (i.e. integrin, adhesion molecule) known to be modulated by FGF-2 and involved in angiogenesis, and by quantifying closed areas delimited by vascular tubes formed on reconstituted basement membrane. Fucoidan had no modulatory effect on the mitogenic activity of FGF-2, but significantly increased tubular structure density induced by FGF-2. Fucoidan alone increased alpha(6) integrin subunit expression with only partially organized tubular structure. In the presence of FGF-2, fucoidan enhanced alpha(6), beta(1) and PECAM-1 and inhibited alpha(v)beta(3) integrin expression. Heparin had no effect in these systems. The most striking effect of fucoidan was observed on alpha(6) expression and tube formation was abolished by monoclonal anti-alpha(6) antibodies. Fucoidan plus FGF-2 effect on alpha(6) expression was markedly decreased by monoclonal anti-FGF-2 antibodies, indicating that fucoidan acts mainly via FGF-2. These results show that, at **antithrombotic** concentrations, contrary to heparin, fucoidan can enhance vascular tube formation induced by FGF-2 with a modulation of the expression of surface proteins (mainly alpha(6)) involved in angiogenesis.

L23 ANSWER 3 OF 36 MEDLINE on STN
ACCESSION NUMBER: 2001084280 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10959709
TITLE: Modulation of vascular human endothelial and rat smooth muscle cell growth by a fucosylated chondroitin **sulfate** from echinoderm.
AUTHOR: Tapon-Bretaudiere J; Drouet B; Matou S; Mourao P A; Bros A; **Letourneur** D; Fischer A M
CORPORATE SOURCE: Laboratoire d'Hematologie, CHU Necker, INSERM U428, Universite Paris V, France.. jacqueline.tapon-bretaudiere@nck.ap-hop-paris.fr
SOURCE: Thrombosis and haemostasis, (2000 Aug) 84 (2) 332-7. Journal code: 7608063. ISSN: 0340-6245.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200101
ENTRY DATE: Entered STN: 20010322
Last Updated on STN: 20010322
Entered Medline: 20010118

AB Fucosylated chondroitin **sulfate** is a glycosaminoglycan extracted from the sea cucumber Ludwigothurea grisea. This **polysaccharide** has the same structure as a mammalian chondroitin **sulfate** but some of the glucuronic acid residues display **sulfated** fucose branches. **Anticoagulant** and **antithrombotic** properties of fucosylated chondroitin **sulfate** have already been described. In order to further investigate its potential therapeutic use as an **antithrombotic** agent, we studied its effect on vascular smooth muscle cell (SMC) proliferation and endothelial cell proliferation, migration and Tissue Factor Pathway Inhibitor (TFPI) release. The experiments were performed on SMC from rat thoracic aorta and on human umbilical vein endothelial cell (HUVEC) in culture with or without added fibroblast growth factors (FGF-1 and FGF-2). Our results showed that: (i) fucosylated chondroitin **sulfate** had a strong inhibitory effect on SMC proliferation (IC50 =10 +/- 5 microg/ml) and (ii) no effect on HUVEC proliferation and migration assays, in the absence of exogenous FGF, while heparin had inhibitory effects; (iii) fucosylated chondroitin **sulfate** (10 microg/ml) enhanced FGF-1 and FGF-2 induced HUVEC proliferation by 45% (145.4 +/- 7.2%) and 27% (126.9 +/- 4.2%), respectively; (iv) on FGF-induced HUVEC migration, fucosylated chondroitin **sulfate** (10 microg/ml) had a strong enhancing effect with FGF-1, +122% (222.2 +/- 15.8%), three times higher than that of heparin, and a lower enhancing effect with FGF-2, +43% (142.7 +/- 4.6%), whereas heparin had no effect; (v) fucosylated chondroitin **sulfate** stimulated TFPI release, mainly on the free form. +98% (198.2 +/- 25%). In addition, the structural features of the **polysaccharide** associated with its biological activity were resolved using chemically modified fucosylated chondroitin **sulfates**. **Sulfated** fucose branches groups are essential to the potentiating effect of the **polysaccharide** on HUVEC proliferation and migration. Surprisingly, removal of fucose branches from the fucosylated chondroitin **sulfate** did not abolish TFPI release. Finally, partial reduction of the glucuronic acid carboxyl groups limited the potentiating effect on HUVEC proliferation and migration but did not affect TFPI release. In conclusion, this fucosylated chondroitin **sulfate** from invertebrate origin reveals useful properties for an

antithrombotic agent: inhibition of SMC proliferation, enhancement of endothelium wound repair and TFPI release. These properties on vascular cells, associated with a low bleeding tendency and an **antithrombotic** activity, strongly suggest its potential use as a new therapeutic agent in arterial **thrombosis** and restenosis, with a more favorable effect than heparin.

L23 ANSWER 4 OF 36 MEDLINE on STN
ACCESSION NUMBER: 1999200590 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10102467
TITLE: **Antithrombotic** and **anticoagulant** activities of a low molecular weight fucoidan by the subcutaneous route.
AUTHOR: Millet J; **Jouault S C**; Mauray S; Theveniaux J; Sternberg C; Boisson Vidal C; Fischer A M
CORPORATE SOURCE: Laboratoires Fournier, Dijon, France.. j.millet@fournier.fr
SOURCE: Thrombosis and haemostasis, (1999 Mar) 81 (3) 391-5. Journal code: 7608063. ISSN: 0340-6245.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199906
ENTRY DATE: Entered STN: 19990714
Last Updated on STN: 19990714
Entered Medline: 19990629

AB Fucoidans (high-molecular-weight **sulfated polysaccharides** extracted from brown seaweeds) have **anticoagulant** and **antithrombotic** effects. They inhibit thrombin by catalyzing both serpins (**antithrombin** and heparin cofactor II) according to their chemical structures and origins. In this study, a low-molecular-weight (LMW) fucoidan of 8 kDa was obtained by chemical degradation of a high-molecular-weight fraction. The **antithrombotic** and **anticoagulant** activities of this new compound were compared to those of a low-molecular-weight heparin (LMWH), dalteparin, following subcutaneous administration to rabbits. This LMW fucoidan exhibited dose-related venous **antithrombotic** activity, with an ED80 of about 20 mg/kg, 2 h after a single subcutaneous injection. Its activity was comparable to that of dalteparin (close to 200 anti-Xa IU/kg) and was maximal 30 min after a single subcutaneous injection. The activity remained stable (about 70%) from 1 to 4 h after injection, but disappeared by 8 h. The **antithrombotic** activity was not associated with either a prolongation of the thrombin clotting time (TCT) or an increase in anti-Xa activity, contrary to dalteparin. A slight prolongation of APTT occurred with both compounds. This venous **antithrombotic** activity was associated with a decrease in ex vivo thrombin generation and with a significant increase in the lag phase in a thrombin generation test. LMW fucoidan thus has potent **antithrombotic** activity and a potentially weaker haemorrhagic effect (i.e. a smaller effect on coagulation tests and a smaller prolongation of the bleeding time) than dalteparin.

L23 ANSWER 5 OF 36 MEDLINE on STN
ACCESSION NUMBER: 1999127850 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9930660
TITLE: Modulation of human endothelial cell proliferation and migration by fucoidan and heparin.
AUTHOR: Giraux J L; Matou S; Bros A; Tapon-Bretaudiere J; **Letourneur D**; Fischer A M
CORPORATE SOURCE: Laboratoire d'Hematologie, Tour Pasteur, Hopital Necker-Enfants Malades, Universite Paris V, France.
SOURCE: European journal of cell biology, (1998 Dec) 77 (4) 352-9. Journal code: 7906240. ISSN: 0171-9335.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199904
ENTRY DATE: Entered STN: 19990426
Last Updated on STN: 19990426
Entered Medline: 19990413

AB Fucoidan is a **sulfated polysaccharide** extracted from brown seaweeds. It has **anticoagulant** and **antithrombotic** properties and inhibits, as well as heparin, vascular smooth muscle cell growth. In this study, we investigated, in the presence of serum and human recombinant growth factors, the effects of fucoidan and heparin on the growth and migration of human umbilical vein endothelial cells (HUVEC) in culture. We found that fucoidan stimulated fetal bovine serum-induced

HUVEC proliferation, whereas heparin inhibited it. In the presence of fibroblast growth factor-1 (FGF-1), both fucoidan and heparin potentiated HUVEC growth. In contrast, fucoidan and heparin inhibited HUVEC proliferation induced by FGF-2, but did not influence the mitogenic activity of vascular endothelial growth factor (VEGF). In the in vitro migration assay from a denuded area of confluent cells, the two **sulfated polysaccharides** markedly enhanced the migration of endothelial cells in the presence of FGF-1. Finally, a weak inhibitory effect on cell migration was found only with the two **polysaccharides** at high concentrations (> or = 100 micro/ml) in presence of serum or combined with FGF-2. All together, the results indicated that heparin and fucoidan can be used as tools to further investigate the cellular mechanisms regulating the proliferation and migration of human vascular cells. Moreover, the data already suggest a potential role of fucoidan as a new therapeutic agent of vegetal origin in the vascular endothelium wound repair.

L23 ANSWER 6 OF 36 MEDLINE on STN
ACCESSION NUMBER: 92245539 MEDLINE
DOCUMENT NUMBER: PubMed ID: 1811335
TITLE: **Anticoagulant** properties of a fucoidan fraction.
AUTHOR: **Colliec S**; Fischer A M; Tapon-Bretaudiere J; Boisson C; Durand P; Jozefonvicz J
CORPORATE SOURCE: CNRS UA502, LRM, C.S.P., Universite Paris Nord, Villetaneuse, France.
SOURCE: Thrombosis research, (1991 Oct 15) 64 (2) 143-54. Journal code: 0326377. ISSN: 0049-3848.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199206
ENTRY DATE: Entered STN: 19920619
Last Updated on STN: 19920619
Entered Medline: 19920603

AB Fucoidans are a family of high molecular weight **sulphated polysaccharides** in the Mr range 8 x 10(5) -10(6), widely dispersed in brown seaweed cell wall. When extracted from several brown algae, they exhibit **anticoagulant** properties. The chemical degradation of a crude extract, from Pelvetia canaliculata, was undertaken to obtain a low molecular weight **polysaccharide** (Mr 20,000 +/- 5,000) with the purpose of a possible clinical use. Its **anticoagulant** potency was investigated through the inhibition of factor IIa and factor Xa in the presence of **antithrombin** III or heparin cofactor II. The degraded fucoidan revealed a potent **antithrombin** activity: studied in an **antithrombin** III depleted plasma or in the presence of purified heparin cofactor II, the fucoidan was as efficient as heparin and dermatan **sulphate** on heparin cofactor II potentiation, at the same concentration by weight. In whole plasma or in the presence of the purified inhibitor, an anti-factor IIa activity mediated by **antithrombin** III was detected (30 times less potent than for heparin, on a weight to weight basis). In contrast, no anti-factor Xa activity was detected in the presence of the degraded fucoidan, under the same experimental conditions. These fucoidans, by-products of alginates preparation in the food and cosmetologic industries, are obtained easily. Thus, they may represent a cheap and easy source of a new type of **anticoagulants**.

L23 ANSWER 7 OF 36 EMBASE
on STN
ACCESSION NUMBER: 2000272654 EMBASE
TITLE: Modulation of vascular human endothelial and rat smooth muscle cell growth by a fucosylated chondroitin **sulfate** from echinoderm.
AUTHOR: Tapon-Bretaudiere J.; Drouet B.; Matou S.; Mourao P.A.S.; Bros A.; **Letourneur D.**; Fischer A.M.
CORPORATE SOURCE: Dr. J. Tapon-Bretaudiere, Laboratoire d'Hematologie, Tour Pasteur, Hopital Necker-Enfants Malades, 149 rue de Sevres, 75743 Paris Cedex 15, France. jacqueline.tapon-bretaudiere@nck.ap-hop-paris.fr
SOURCE: Thrombosis and Haemostasis, (2000) 84/2 (332-337). Refs: 47
ISSN: 0340-6245 CODEN: THHADQ
COUNTRY: Germany
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
025 Hematology
030 Pharmacology

037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Fucosylated chondroitin **sulfate** is a glycosaminoglycan extracted from the sea cucumber Ludwigothurea grisea. This **polysaccharide** has the same structure as a mammalian chondroitin **sulfate** but some of the glucuronic acid residues display **sulfated** fucose branches. **Anticoagulant** and **antithrombotic** properties of fucosylated chondroitin **sulfate** have already been described. In order to further investigate its potential therapeutic use as an **antithrombotic** agent, we studied its effect on vascular smooth muscle cell (SMC) proliferation and endothelial cell proliferation, migration and Tissue Factor Pathway Inhibitor (TFPI) release. The experiments were performed on SMC from rat thoracid aorta and on human umbilical vein endothelial cell (HUVEC) in culture with or without added fibroblast growth factors (FGF-1 and FGP-2). Our results showed that: (i) fucosylated chondroitin **sulfate** had a strong inhibitory effect on SMC proliferation (IC50 = 10 ± 5 μg/ml) and (ii) no effect on HUVEC proliferation and migration assays, in the absence of exogenous FGF, while heparin had inhibitory effects; (iii) fucosylated chondroitin **sulfate** (10 μg/ml) enhanced FGF-1 and FGF-2 induced HUVEC proliferation by 45% (145.4 ± 7.2%) and 27% (126.9 ± 4.2%), respectively; (iv) on FGF-induced HUVEC migration, fucosylated chondroitin **sulfate** (10 μg/ml) had a strong enhancing effect with FGF-1, +122% (222.2 ± 15.8%), three times higher than that of heparin, and a lower enhancing effect with FGF-2, +43% (142.7 ± 4.6%), whereas heparin had no effect; (v) fucosylated chondroitin **sulfate** stimulated TFPI release, mainly on the free form, +98% (198.2 ± 25.%). In addition, the structural features of the **polysaccharide** associated with its biological activity were resolved using chemically modified fucosylated chondroitin **sulfates**. **Sulfated** fucose branches groups are essential to the potentiating effect of the **polysaccharide** on HUVEC proliferation and migration. Surprisingly, removal of fucose branches from the fucosylated chondroitin **sulfate** did not abolish TFPI release. Finally, partial reduction of the glucuronic acid carboxyl groups limited the potentiating effect on HUVEC proliferation and migration but did not affect TFPI release. In conclusion, this fucosylated chondroitin **sulfate** from invertebrate origin reveals useful properties for an **antithrombotic** agent: inhibition of SMC proliferation, enhancement of endothelium wound repair and TFPI release. These properties on vascular cells, associated with a low bleeding tendency and an **antithrombatic** activity, strongly suggest its potential use as a new therapeutic agent in arterial **thrombosis** and restenosis, with a more favorable effect than heparin.

L23 ANSWER 8 OF 36 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
ACCESSION NUMBER: 1999090358 EMBASE
TITLE: **Antithrombotic** and **anticoagulant** activities of a low molecular weight fucoidan by the subcutaneous route.
AUTHOR: Millet J.; **Jouault S.C.**; Mauray S.; Theveniaux J.; Sternberg C.; Boisson Vidal C.; Fischer A.M.
CORPORATE SOURCE: Dr. J. Millet, Laboratoires Fournier, 50 rue de Dijon, Daix, France. j.millet@fournier.fr
SOURCE: Thrombosis and Haemostasis, (1999) 81/3 (391-395).
Refs: 28
ISSN: 0340-6245 CODEN: THHADQ
COUNTRY: Germany
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 025 Hematology
030 Pharmacology
037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Fucoidans (high-molecular-weight **sulfated** **polysaccharides** extracted from brown seaweeds) have **anticoagulant** and **antithrombotic** effects. They inhibit thrombin by catalyzing both serpins (**antithrombin** and heparin cofactor II) according to their chemical structures and origins. In this study, a low-molecular-weight (LMW) fucoidan of 8 kDa was obtained by chemical degradation of a high-molecular-weight fraction. The **antithrombotic** and **anticoagulant** activities of this new compound were compared to those of a low-molecular-weight heparin (LMWH), dalteparin, following subcutaneous administration to rabbits. This LMW fucoidan exhibited dose-related venous **antithrombotic** activity, with an ED80 of about 20 mg/kg, 2 h after a single subcutaneous injection.

Its activity was comparable to that of dalteparin (close to 200 anti-Xa IU/kg) and was maximal 30 min after a single subcutaneous injection. The activity remained stable (about 70%) from 1 to 4 h after injection, but disappeared by 8 h. The **antithrombotic** activity was not associated with either a prolongation of the thrombin clotting time (TCT) or an increase in anti-Xa activity, contrary to dalteparin. A slight prolongation of APTT occurred with both compounds. This venous **antithrombotic** activity was associated with a decrease in ex vivo thrombin generation and with a significant increase in the lag phase in a thrombin generation test. LMW fucoidan thus has potent **antithrombotic** activity and a potentially weaker haemorrhagic effect (i.e. a smaller effect on coagulation tests and a smaller prolongation of the bleeding time) than dalteparin.

L23 ANSWER 9 OF 36 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN

ACCESSION NUMBER: 1999013235 EMBASE
TITLE: Modulation of human endothelial cell proliferation and migration by fucoidan and heparin.
AUTHOR: Giraux J.-L.; Matou S.; Bros A.; Tapon-Bretaudiere J.; **Letourneur D.**; Fischer A.-M.
CORPORATE SOURCE: Prof. A.-M. Fischer, Laboratoire d'Hematologie, Hopital Necker-Enfants Malades, Universite Paris V, 149 rue de Sevres, F-75743 Paris, France. anne-marie.fischer@nck.ap-hop-paris.fr
SOURCE: European Journal of Cell Biology, (1998) 77/4 (352-359). Refs: 39
ISSN: 0171-9335 CODEN: EJCBDN
COUNTRY: Germany
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Fucoidan is a **sulfated polysaccharide** extracted from brown seaweeds. It has **anticoagulant** and **antithrombotic** properties and inhibits, as well as heparin, vascular smooth muscle eel growth. In this study, we investigated, in the presence of serum and human recombinant growth factors, the effects of fucoidan and heparin on the growth and migration of human umbilical vein endothelial cells (HUVEC) in culture. We found that fucoidan stimulated fetal bovine serum-induced HUVEC proliferation, whereas heparin inhibited it. In the presence of fibroblast growth factor-1 (FGF-1), both fucoidan and heparin potentiated HUVEC growth. In contrast, fucoidan and heparin inhibited HUVEC proliferation induced by FGF-2, but did not influence the mitogenic activity of vascular endothelial growth factor (VEGF). In the in vitro migration assay from a denuded area of confluent cells, the two **sulfated polysaccharides** markedly enhanced the migration of endothelial cells in the presence of FGF-1. Finally a weak inhibitory effect on cell migration was found only with the two **polysaccharides** at high concentrations (≥ 100 μg/ml) in presence of serum or combined with FGF-2. All together, the results indicated that heparin and fucoidan can be used as tools to further investigate the cellular mechanisms regulating the proliferation and migration of human vascular cells. Moreover, the data already suggest a potential role of fucoidan as a new therapeutic agent of vegetal origin in the vascular endothelium wound repair.

L23 ANSWER 10 OF 36 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN

ACCESSION NUMBER: 92009626 EMBASE
DOCUMENT NUMBER: 1992009626
TITLE: **Anticoagulant** properties of a fucoidan fraction.
AUTHOR: **Colliec S.**; Fischer A.M.; Tapon-Bretaudiere J.; Boisson C.; Durand P.; Jozefonvicz J.
CORPORATE SOURCE: Laboratoire d'Hematologie, C.H.U Necker-Enfants Malades, 156 Rue de Vaugirard,75730 Paris Cedex 15, France
SOURCE: Thrombosis Research, (1991) 64/2 (143-154).
ISSN: 0049-3848 CODEN: THBRAA
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 025 Hematology
037 Drug Literature Index
030 Pharmacology
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Fucoidans are a family of high molecular weight **sulphated polysaccharides** in the Mr range 8 x 105 - 106, widely dispersed in brown seaweed cell wall. When extracted from several brown algae, they

exhibit **anticoagulant** properties. The chemical degradation of a crude extract, from Pelvetia canaliculata, was undertaken to obtain a low molecular weight **polysaccharide** (Mr 20,000 ± 5,000) with the purpose of a possible clinical use. Its **anticoagulant** potency was investigated through the inhibition of factor IIa and factor Xa in the presence of **antithrombin** III or heparin cofactor II. The degraded fucoidan revealed a potent **antithrombin** activity: studied in an **antithrombin** II depleted plasma or in the presence of purified heparin cofactor II, the fucoidan was as efficient as heparin and dermatan **sulphate** on heparin cofactor II potentiation, at the same concentration by weight. In whole plasma or in the presence of the purified inhibitor, an anti-factor IIa activity mediated by **antithrombin** III was detected (30 times less potent than for heparin, on a weight to weight basis). In contrast, no anti-factor Xa activity was detected in the presence of the degraded fucoidan, under the same experimental conditions. These fucoidans, by-products of alginates preparation in the food and cosmetologic industries, are obtained easily. Thus, they may represent a cheap and easy source of a new type of **anticoagulants**.

L23 ANSWER 11 OF 36 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2004:49891 BIOSIS
DOCUMENT NUMBER: PREV200400053590
TITLE: Interactions of heparin with human skin cells: Binding, location, and transdermal penetration.
AUTHOR(S): Parisel, Claire; Saffar, Line; Gattegno, Liliane; Andre, Valerie; Abdul-Malak, Nabil; Perrier, Eric; **Letourneur, Didier** [Reprint Author]
CORPORATE SOURCE: INSERM ERIT-M 0204, X. Bichat Hospital, University Paris VII and University Paris XIII, 75877, Paris Cedex, 18, France
didier.letourneur@galilee.univ-paris13.fr
SOURCE: Journal of Biomedical Materials Research, (November 1 2003) Vol. 67A, No. 2, pp. 517-523. print.
ISSN: 0021-9304 (ISSN print).
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 21 Jan 2004
Last Updated on STN: 21 Jan 2004

AB The development of new materials for tissue engineering of skin substitutes requires an increasing knowledge of their interactions with human skin cells. Since carbohydrate recognition is involved in numerous biologic processes, including skin regeneration, the aim of this study was to identify sugar receptors expressed at the surface of human dermic and epidermic cells. Binding of fluorescent sugar-polyhydroxyethylacrylamide derivatives was analyzed by flow cytofluorimetry on cultured human skin fibroblasts, keratinocytes, and melanocytes. We observed that these three cell types express a membrane receptor specific for GlcNAc6S. Since the **polysaccharide** heparin contains this sugar moiety, we further investigated the interactions of heparin with skin cells. We analyzed the in vitro cell binding and ex vivo diffusion with the Franz cell of heparin and of two other **polysaccharides** of similar molecular weight, dextran and chondroitin **sulfate**. We found evidence of the preferential binding of heparin on keratinocytes and its high transcutaneous penetration of skin. Altogether, our results describe the affinity of heparin for human skin cells and suggest it may be an excellent candidate for use in the skin delivery of drugs or cosmetics and also as an active component in engineered skin.

L23 ANSWER 12 OF 36 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2003:10136 BIOSIS
DOCUMENT NUMBER: PREV200300010136
TITLE: Effect of fucoidan on fibroblast growth factor-2-induced angiogenesis in vitro.
AUTHOR(S): Matou, Sabine; Helley, Dominique [Reprint Author]; Chabut, Delphine; Bros, Andree; **Fischer, Anne-Marie**
CORPORATE SOURCE: Hopital Europeen Georges Pompidou, 20 Rue Leblanc, 75908, Paris Cedex 15, France
dominique.helley@egp.ap-hop-paris.fr
SOURCE: Thrombosis Research, (May 15 2002) Vol. 106, No. 4-5, pp. 213-221. print.
CODEN: THBRAA. ISSN: 0049-3848.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 18 Dec 2002
Last Updated on STN: 18 Dec 2002

AB Fucoidans are **sulfated polysaccharides** extracted from brown marine algae. A purified fucoidan fraction exhibits the same venous

antithrombotic activity as heparin in rabbits, but with a lower **anticoagulant** effect. Because of its heparin-like structure, we postulated that fucoidan might modulate heparin-binding angiogenic growth factor activity. We thus studied its effect, at **antithrombotic** concentrations, on fibroblast growth factor (FGF)-2-induced proliferation and differentiation of human umbilical vein endothelial cells. The fucoidan effect on endothelial cell differentiation was evaluated by studying the expression of surface proteins (i.e. integrin, adhesion molecule) known to be modulated by FGF-2 and involved in angiogenesis, and by quantifying closed areas delimited by vascular tubes formed on reconstituted basement membrane. Fucoidan had no modulatory effect on the mitogenic activity of FGF-2, but significantly increased tubular structure density induced by FGF-2. Fucoidan alone increased alpha6 integrin subunit expression with only partially organized tubular structure. In the presence of FGF-2, fucoidan enhanced alpha6, beta1 and PECAM-1 and inhibited alphavbeta3 integrin expression. Heparin had no effect in these systems. The most striking effect of fucoidan was observed on alpha6 expression and tube formation was abolished by monoclonal anti-alpha6 antibodies. Fucoidan plus FGF-2 effect on alpha6 expression was markedly decreased by monoclonal anti-FGF-2 antibodies, indicating that fucoidan acts mainly via FGF-2. These results show that, at **antithrombotic** concentrations, contrary to heparin, fucoidan can enhance vascular tube formation induced by FGF-2 with a modulation of the expression of surface proteins (mainly alpha6) involved in angiogenesis.

L23 ANSWER 13 OF 36 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2001:564167 BIOSIS
DOCUMENT NUMBER: PREV200100564167
TITLE: Characterization, chemical modifications and in vitro **anticoagulant** properties of an exopolysaccharide produced by Alteromonas infernus.
AUTHOR(S): **Jouault, Sylvia Colliec** [Reprint author]; Chevolot, Lionel; Helley, Dominique; Ratiskol, Jacqueline; Bros, Andree; Sinquin, Corinne; Roger, Olivier; **Fischer, Anne-Marie**
CORPORATE SOURCE: Laboratoire de Biochimie et Molecules Marines, Departement Valorisation des Produits, URM2, IFREMER/CNRS (UMR 7540, CNRS/Universite Paris 13), 44311, Nantes Cedex 3, France sylvia.colliec.jouault@ifremer.fr
SOURCE: Biochimica et Biophysica Acta, (3 October, 2001) Vol. 1528, No. 2-3, pp. 141-151. print.
CODEN: BBACAQ. ISSN: 0006-3002.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 5 Dec 2001
Last Updated on STN: 25 Feb 2002

AB A new low-molecular-weight 'heparin-like' component was obtained from an exopolysaccharide produced by a mesophilic strain found in deep-sea hydrothermal vents. Data concerning the structure of the native high-molecular-weight exopolysaccharide (106 g/mol, 10% **sulfate** content) are reported for the first time. Two depolymerization processes were used to obtain low-molecular-weight (24-35X103 g/mol) oversulfated fractions (**sulfate** content 20 or 40%). Nuclear magnetic resonance studies indicated that after **sulfation** (40%), the low-molecular-weight fraction obtained by free radical depolymerization was less **sulfated** in the 6-O-position than the fraction depolymerized by acid hydrolysis. The free radical depolymerized product also had **sulfated** residues in the 4-O-position and disulfated ones in the 2,3-O-positions. Moreover, the compounds generated by the free radical process were more homogeneous with respect to molecular mass. Also for the first time, the **anticoagulant** activity of the low-molecular-weight exopolysaccharide fractions is reported. When the fractions obtained after **sulfation** and depolymerization were compared with heparins, **anticoagulant** activity was detected in oversulfated fractions, but not in native exopolysaccharide. The free radical depolymerized fraction inhibited thrombin generation in both contact-activated and thromboplastin-activated plasma, showing a prolonged lag phase only in the contact-activated assay. Affinity co-electrophoresis studies suggested that a single population of **polysaccharide** chains binds to **antithrombin** and that only a subpopulation strongly interacts with heparin cofactor II.

L23 ANSWER 14 OF 36 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2001:273907 BIOSIS
DOCUMENT NUMBER: PREV200100273907
TITLE: Inactivation of thrombin by a fucosylated chondroitin **sulfate** from echinoderm.
AUTHOR(S): Mourao, Paulo A. S. [Reprint author]; Boisson-Vidal,

Catherine; Tapon-Bretaudiere, Jacqueline; Drouet, Bruno; Bros, Andree; **Fischer, Anne-Marie**

CORPORATE SOURCE: Laboratorio de Tecido Conjuntivo, Hospital Universitario Clementino Fraga Filho and Departamento de Bioquimica Medica, Centro de Ciencias da Saude, Universidade Federal do Rio de Janeiro, Rio de Janeiro, RJ, 21941-590, Brazil pmourao@hucff.ufrj.br

SOURCE: Thrombosis Research, (April 15, 2001) Vol. 102, No. 2, pp. 167-176. print.
CODEN: THBRAA. ISSN: 0049-3848.

DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 6 Jun 2001
Last Updated on STN: 19 Feb 2002

AB A **polysaccharide** extracted from the sea cucumber body wall has the same backbone structure as the mammalian chondroitin **sulfate**, but some of the glucuronic acid residues display **sulfated** fucose branches. These branches confer high **anticoagulant** activity to the **polysaccharide**. Since the sea cucumber chondroitin **sulfate** has analogy in structure with mammalian glycosaminoglycans and **sulfated fucans** from brown algae, we compared its **anticoagulant** action with that of heparin and of a homopolymeric **sulfated fucan** with approximately the same level of **sulfation** as the **sulfated** fucose branches found in the sea cucumber **polysaccharide**. These various compounds differ not only in their **anticoagulant** potencies but also in the mechanisms of thrombin inhibition. Fucosylated chondroitin **sulfate**, like heparin, requires **antithrombin** or heparin cofactor II for thrombin inhibition. **Sulfated fucans** from brown algae have an **antithrombin** effect mediated by **antithrombin** and heparin cofactor II, plus a direct **antithrombin** effect more pronounced for some fractions. But even in the case of these two **polysaccharides**, we observed some differences. In contrast with heparin, total inhibition of thrombin in the presence of **antithrombin** is not achieved with fucosylated chondroitin **sulfate**, possibly reflecting a less specific interaction. Fucosylated chondroitin **sulfate** is able to inhibit thrombin generation after stimulation by both contact-activated and thromboplastin-activated systems. It delayed only the contact-induced thrombin generation, as expected for an **anticoagulant** without direct thrombin inhibition. Overall, the specific spatial array of the **sulfated** fucose branches in the fucosylated chondroitin **sulfate** not only confer high **anticoagulant** activity to the **polysaccharide** but also determine differences in the way it inhibits thrombin.

L23 ANSWER 15 OF 36 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2001:159748 BIOSIS
DOCUMENT NUMBER: PREV200100159748
TITLE: Relationship between **antithrombotic** activities of **fucans** and their structure.

AUTHOR(S): Boisson-Vidal, Catherine [Reprint author]; Chaubet, Frederic; Chevolot, Lionel; Sinquin, Corinne; Theveniaux, Jocelyne; **Millet, Jean**; Sternberg, Claude; Mulloy, Barbara; **Fischer, Anne Marie**

CORPORATE SOURCE: Laboratoire de Recherches en Hemostate, Hopital Necker-Enfants Malades, Paris, France cathbv@galilee.univ-paris13.fr

SOURCE: Drug Development Research, (December, 2000) Vol. 51, No. 4, pp. 216-224. print.
CODEN: DDREDK. ISSN: 0272-4391.

DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 28 Mar 2001
Last Updated on STN: 15 Feb 2002

AB A low molecular weight **fucan** fraction extracted from the brown seaweed Ascophyllum nodosum was previously shown to exhibit dose-related venous **antithrombotic** activity with an ED80 of about 20 mg/kg, 2 h after a single subcutaneous injection HCII (Colliec et al. (1991) Thromb Res 64:143-154; Mauray et al. (1995) Thromb Haemast 74:1280-1285). Its activity was comparable to that of a low molecular weight heparin (Dalteparin(R)). This **fucan** fraction is one of several, with a range of different structure parameters, prepared by degradation of the whole native **fucan**. These low molecular weight fractions were compared using a Wessler stasis **thrombosis** model in rabbits and by determination of their in vitro and ex vivo **anticoagulant** activities. Intravenous administrations of these fractions reduced

thrombosis in a dose-dependent manner. Partial removal of **sulfate** groups and/or partial degradation lead to a significant decrease in their **anticoagulant** and **antithrombotic** activities. The integrity of the regular pattern of **sulphation** of the fucoidan is necessary for **antithrombotic** activity.

L23 ANSWER 16 OF 36 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 2000:414718 BIOSIS
DOCUMENT NUMBER: PREV200000414718
TITLE: Modulation of vascular human endothelial and rat smooth muscle cell growth by a fucosylated chondroitin **sulfate** from echinoderm.
AUTHOR(S): Tapon-Bretaudiere, J. [Reprint author]; Drouet, B.; Matou, S.; Mourao, P. A. S.; Bros, A.; **Letourneur, D.**; Fischer, A. M.
CORPORATE SOURCE: Laboratoire d'Hematologie, Tour Pasteur, Hopital Necker-Enfants Malades, 149 rue de Sevres, 75743, Paris Cedex, 15, France
SOURCE: Thrombosis and Haemostasis, (August, 2000) Vol. 84, No. 2, pp. 332-337. print.
CODEN: THHADQ. ISSN: 0340-6245.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 27 Sep 2000
Last Updated on STN: 8 Jan 2002

AB Fucosylated chondroitin **sulfate** is a glycosaminoglycan extracted from the sea cucumber Ludwigothurea grisea. This **polysaccharide** has the same structure as a mammalian chondroitin **sulfate** but some of the glucuronic acid residues display **sulfated** fucose branches. **Anticoagulant** and **antithrombotic** properties of fucosylated chondroitin **sulfate** have already been described. In order to further investigate its potential therapeutic use as an **antithrombotic** agent, we studied its effect on vascular smooth muscle cell (SMC) proliferation and endothelial cell proliferation, migration and Tissue Factor Pathway Inhibitor (TFPI) release. The experiments were performed on SMC from rat thoracic aorta and on human umbilical vein endothelial cell (HUVEC) in culture with or without added fibroblast growth factors (FGF-1 and FGF-2). Our results showed that: (i) fucosylated chondroitin **sulfate** had a strong inhibitory effect on SMC proliferation (IC50 = 10 +- 5 mug/ml) and (ii) no effect on HUVEC proliferation and migration assays, in the absence of exogenous FGF, while heparin had inhibitory effects; (iii) fucosylated chondroitin **sulfate** (10 mug/ml) enhanced FGF-1 and FGF-2 induced HUVEC proliferation by 45% (145.4 +- 7.2%) and 27% (126.9 +- 4.2%), respectively; (iv) on FGF-induced HUVEC migration, fucosylated chondroitin **sulfate** (10 mug/ml) had a strong enhancing effect with FGF-1, +122% (222.2 +- 15.8%), three times higher than that of heparin, and a lower enhancing effect with FGF-2, +43% (142.7 +- 4.6%), whereas heparin had no effect; (v) fucosylated chondroitin **sulfate** stimulated TFPI release, mainly on the free form, +98% (198.2 +- 25.%). In addition, the structural features of the **polysaccharide** associated with its biological activity were resolved using chemically modified fucosylated chondroitin **sulfates**. **Sulfated** fucose branches groups are essential to the potentiating effect of the **polysaccharide** on HUVEC proliferation and migration. Surprisingly, removal of fucose branches from the fucosylated chondroitin **sulfate** did not abolish TFPI release. Finally, partial reduction of the glucuronic acid carboxyl groups limited the potentiating effect on HUVEC proliferation and migration but did not affect TFPI release. In conclusion, this fucosylated chondroitin **sulfate** from invertebrate origin reveals useful properties for an **antithrombotic** agent: inhibition of SMC proliferation, enhancement of endothelium wound repair and TFPI release. These properties on vascular cells, associated with a low bleeding tendency and an **antithrombotic** activity, strongly suggest its potential use as a new therapeutic agent in arterial **thrombosis** and restenosis, with a more favorable effect than heparin.

L23 ANSWER 17 OF 36 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1999:188014 BIOSIS
DOCUMENT NUMBER: PREV199900188014
TITLE: Modulation of human endothelial cell proliferation and migration by fucoidan and heparin.
AUTHOR(S): Giraux, Jean-Luc; Matou, Sabine; Bros, Andree; Tapon-Bretaudiere, Jacqueline; **Letourneur, Didier** ; **Fischer, Anne-Marie** [Reprint author]
CORPORATE SOURCE: Laboratoire d'Hematologie, Tour Pasteur, Hopital Necker-Enfants Malades, Universite Paris V, 149, rue de

Sevres, F-75743, Paris Cedex 15, France
SOURCE: European Journal of Cell Biology, (Dec., 1998) Vol. 77, No. 4, pp. 352-359. print.
CODEN: EJCBDN. ISSN: 0171-9335.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 5 May 1999
Last Updated on STN: 5 May 1999

AB Fucoidan is a **sulfated polysaccharide** extracted from brown seaweeds. It has **anticoagulant** and **antithrombotic** properties and inhibits, as well as heparin, vascular smooth muscle cell growth. In this study, we investigated, in the presence of serum and human recombinant growth factors, the effects of fucoidan and heparin on the growth and migration of human umbilical vein endothelial cells (HUVEC) in culture. We found that fucoidan stimulated fetal bovine serum-induced HUVEC proliferation, whereas heparin inhibited it. In the presence of fibroblast growth factor-1 (FGF-1), both fucoidan and heparin potentiated HUVEC growth. In contrast, fucoidan and heparin inhibited HUVEC proliferation induced by FGF-2, but did not influence the mitogenic activity of vascular endothelial growth factor (VEGF). In the in vitro migration assay from a denuded area of confluent cells, the two **sulfated polysaccharides** markedly enhanced the migration of endothelial cells in the presence of FGF-1. Finally, a weak inhibitory effect on cell migration was found only with the two **polysaccharides** at high concentrations (>EQ;100 mug/ml) in presence of serum or combined with FGF-2. All together, the results indicated that heparin and fucoidan can be used as tools to further investigate the cellular mechanisms regulating the proliferation and migration of human vascular cells. Moreover, the data already suggest a potential role of fucoidan as a new therapeutic agent of vegetal origin in the vascular endothelium wound repair.

L23 ANSWER 18 OF 36 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1998:500333 BIOSIS
DOCUMENT NUMBER: PREV199800500333
TITLE: Fucoidan, as heparin, induces tissue factor pathway inhibitor release from cultured human endothelial cells.
AUTHOR(S): Giraux, Jean-Luc; Tapon-Bretaudiere, Jacqueline; Matou, Sabine; **Fischer, Anne-Marie** [Reprint author]
CORPORATE SOURCE: Lab. d'Hematol., Tour Pasteur, Hopital Necker-Enfants Malades, 149 rue de Sevres, 75743 Paris Cedex 15, France
SOURCE: Thrombosis and Haemostasis, (Oct., 1998) Vol. 80, No. 4, pp. 692-695. print.
CODEN: THHADQ. ISSN: 0340-6245.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 18 Nov 1998
Last Updated on STN: 18 Nov 1998

AB Fucoidan, a **sulfated polysaccharide** extracted from brown seaweeds, has **antithrombotic** properties, the mechanism of which is not yet completely understood. Tissue factor pathway inhibitor (TFPI), which regulates the tissue factor-dependent pathway of blood coagulation, is released from the endothelium by heparin, a mechanism contributing to its **antithrombotic** activity. In this study, we demonstrated that fucoidan, as heparin, induces TFPI release from cultured human umbilical vein endothelial cells (HUVEC). The TFPI accumulation in the HUVEC supernatants depends on the incubation time and **polysaccharide** concentration. After 30 to 60 minutes of incubation, TFPI concentration (total antigen level) was twice higher in the presence of both **polysaccharides** than in their absence. After one hour of incubation, in the presence of increasing concentrations of each **polysaccharide**, an optimal stimulation was observed for 0.5 mug/ml of fucoidan and 5 mug/ml of heparin, as evidenced by a raise of the basal TFPI level: a 2-fold increase for the total antigen and a 3-fold increase for the free antigen. These data suggest that TFPI released from vascular-endothelial cells may contribute to the **antithrombotic** effect of fucoidan.

L23 ANSWER 19 OF 36 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1998:485412 BIOSIS
DOCUMENT NUMBER: PREV199800485412
TITLE: Mechanism of factor IXa inhibition by **antithrombin** in the presence of unfractionated and low molecular weight heparins and fucoidan.
AUTHOR(S): Mauray, Sandrine; De Raucourt, Emmanuelle; Talbot, Jean-Claude; Dachary-Prigent, Jeanne; Jozefowicz, Marcel; **Fischer, Anne-Marie** [Reprint author]
CORPORATE SOURCE: Laboratoire Recherche Hematologie, Hopital Necker

Enfants-Malades, Universite Paris V, 75743 Paris Cedex 15, France
SOURCE: Biochimica et Biophysica Acta, (Sept. 8, 1998) Vol. 1387, No. 1-2, pp. 184-194. print.
CODEN: BBACAQ. ISSN: 0006-3002.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 5 Nov 1998
Last Updated on STN: 5 Nov 1998

AB Heparin exerts its **anticoagulant** activity by catalysing the inhibition of coagulation proteases by **antithrombin** (AT). Its main target is thrombin but it also catalyses the inhibition of the other serine-proteases of the coagulation cascade, such as factor IXa (fIXa). The aim of this study was to compare the catalysis of inhibition of blood fIXa by **antithrombin** in the presence of several **sulfated polysaccharides** with **anticoagulant** activity, i.e. heparin, three widely used in therapeutics low molecular weight heparins (LMWH) and fucoidan. Plots of the second-order rate constants of the fIXa-**antithrombin** reaction vs. the concentration of added heparin and LMWH are bell-shaped and fit the kinetic model established for thrombin-**antithrombin** reaction by Jordan R., Beeler D., Rosenberg R. (1979) J. Biol. Chemical, 254, 2902-2913. In the ascending branch, the catalyst (C) binds quickly to the inhibitor (I) to form a catalyst-inhibitor (CI) complex which is more reactive towards the enzyme (E) than the free inhibitor, leading to the formation of an inactive enzyme-inhibitor complex (EI) and the release of free catalyst, in a rate-limiting second step. After a maximum corresponding to an optimal catalyst concentration, the decrease in the reaction rate was in keeping with the formation of a catalyst-enzyme (CE) complex, whose inactivation by the CI complex was slower than that of the free enzyme. Maximum second-order rate constants for the inhibition of fIXa by AT were 105, 6.8, 12.24 and 22 muM-1 min-1 with heparin, Enoxaparin, Fraxiparin and Fragmin, respectively, leading to 3500-, 225-, 405- and 728-fold increases in the inhibition rate in the absence of **polysaccharide**, respectively. Fucoidan yielded 23-fold increase in the fIXa-**antithrombin** interaction rate. The kinetic profiles obtained with this **polysaccharide** exhibited ascending branch which correlated well with the kinetic model based on the formation of binary complexes (CI or CE). Fucoidan was covalently conjugated with a fluorescent probe (DTAF) and used in conjunction with fluorescence anisotropy to follow its binding to **antithrombin**, heparin cofactor II (HCII), thrombin and fIXa. The binding of fucoidan to these proteins occurred with low affinities when compared to heparin and LMWH. Fucoidan had higher affinityfor the inhibitor HCII compared to **antithrombin** and enzymes. These data suggest that binding of heparins and fucoidan to the inhibitor (CI) is required for the **polysaccharide**-dependent enhancement in the rate of neutralization of the enzyme by the inhibitor.

L23 ANSWER 20 OF 36 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1996:440203 BIOSIS
DOCUMENT NUMBER: PREV199699162559
TITLE: Pharmacologic and biochemical profiles of new venous **antithrombotic** beta-D-xyloside derivatives: Potential antiathero/thrombotic drugs.
AUTHOR(S): Martin, Niall B. [Reprint author]; Masson, Philippe; Sepulchre, Christiane; Theveniaux, Jocelyne; **Millet, Jean**; Bellamy, Francois
CORPORATE SOURCE: Laboratoires Fournier, Centre Recherche, 50 rue Dijon, 21121 Daix, France
SOURCE: Seminars in Thrombosis and Hemostasis, (1996) Vol. 22, No. 3, pp. 247-254.
CODEN: STHMBV. ISSN: 0094-6176.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 7 Oct 1996
Last Updated on STN: 5 Nov 1996

L23 ANSWER 21 OF 36 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1995:82470 BIOSIS
DOCUMENT NUMBER: PREV199598096770
TITLE: The venous **antithrombotic** profile of naroparcil in the rabbit.
AUTHOR(S): **Millet, Jean** [Reprint author]; Theveniaux, Jocelyne; Brown, Neil L.
CORPORATE SOURCE: Lab. Fournier S.C.A., Recherche et Developpement, 50 rue de Dijon, F-21121 Daix, France
SOURCE: Thrombosis and Haemostasis, (1994) Vol. 72, No. 6, pp. 874-879.

CODEN: THHADQ. ISSN: 0340-6245.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 22 Feb 1995
Last Updated on STN: 27 Apr 1995

AB The venous **antithrombotic** profile of naroparcil or (4-(4-cyanobenzoyl)-phenyl)-1.5-dithio-beta-D-xylopyranoside was investigated in the rabbit following single i. v. and oral administration. Naroparcil attenuated thrombus development in a Wessler stasis model of venous **thrombosis** (jugular vein) employing bovine factor Xa as a thrombogenic stimulus giving ED-50 values of 21.9 mg/kg and 36.0 mg/kg after respectively i. v. and oral administration. Venous **antithrombotic** activity was maximal 2-3 h after i. v. administration and 4-8 h after oral administration. Four hours after the oral administration of maximal **antithrombotic** (Wessler model, factor Xa) doses (100 and 400 mg/kg), naroparcil had no significant effect on bleeding time. in platelet poor plasma obtained from animals treated 4 h previously with various doses (25 to 400 mg/kg) of naroparcil, there was no detectable anti-factor Xa nor **antithrombin** activity. Similarly, naroparcil had no effect on APTT nor on thrombin time. A sensitized thrombin time (to about 35 s) was modestly but significantly increased following oral administration of the compound at 400 mg/kg. However, thrombin generation by the intrinsic pathway was reduced in a dose-related manner, maximal reduction being 65% at 400 mg/kg. The same doses of naroparcil enhanced the formation of thrombin/heparin cofactor II complexes at the expense of thrombin/**antithrombin** III complexes in plasma incubated with (125I)-human alpha-thrombin and induced the appearance of dermatan **sulfate**-like material in the plasma of treated rabbits, as measured by a heparin cofactor II-mediated thrombin inhibition assay. The results suggest that naroparcil could have a safe venous **antithrombotic** profile following oral administration (**antithrombotic** effect compared to bleeding risk). It is probable that part of the mechanism of action of the beta-D-xyloside, naroparcil, is due to the induction of chondroitin **sulfate**-like glycosaminoglycan biosynthesis, this material being detectable in the plasma.

L23 ANSWER 22 OF 36 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
ACCESSION NUMBER: 1992:75973 BIOSIS
DOCUMENT NUMBER: PREV199293044428; BA93:44428
TITLE: **ANTICOAGULANT** PROPERTIES OF A FUCOIDAN FRACTION.
AUTHOR(S): **COLLIEC S** [Reprint author]; FISCHER A M; TAPON-BRETAUDIERE J; BOISSON C; DURAND P; JOZEFONVICZ J
CORPORATE SOURCE: CNRS UA502, LRM, CSP, UNIV PARIS NORD, 93430 VILLETANEUSE, FR
SOURCE: Thrombosis Research, (1991) Vol. 64, No. 2, pp. 143-154. CODEN: THBRAA. ISSN: 0049-3848.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH
ENTRY DATE: Entered STN: 2 Feb 1992
Last Updated on STN: 2 Feb 1992

AB Fucoidans are a family of high molecular weight **sulphated polysaccharides** in the Mr range 8 + 105 - 106, widely dispersed in brown seaweed cell wall. When extracted from several brown algae, they exhibit **anticoagulant** properties. The chemical degradation of a crude extract, from Pelvetia canaliculata, was undertaken to obtain a low molecular weight **polysaccharide** (Mr 20,000 ± 5,000) with the purpose of a possible clinical use. Its **anticoagulant** potency was investigated through the inhibition of factor IIa and factor Xa in the presence of **antithrombin** III or heparin cofactor II. The degraded fucoidan revealed a potent **antithrombin** activity: studied in an **antithrombin** III depleted plasma or in the presence of purified heparin cofactor II, the fucoidan was as efficient as heparin and dermatan **sulphate** on heparin cofactor II potentiation, at the same concentration by weight. In whole plasma or in the presence of the purified inhibitor, an anti-factor IIa activity mediated by **antithrombin** III was detected (30 times less potent than for heparin, on a weight to weight basis). In contrast, no anti-factor Xa activity was detected in the presence of the degraded fucoidan, under the same experimental conditions. These fucoidans, by-products of alginates preparation in the food and cosmetologic industries, are obtained easily. Thus, they may represent a cheap and easy source of a new type of **anticoagulants**.

L23 ANSWER 23 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2002:724866 CAPLUS
DOCUMENT NUMBER: 138:348472

TITLE: Effect of fucoidan on fibroblast growth factor-2-induced angiogenesis in vitro
AUTHOR(S): Matou, Sabine; Helley, Dominique; Chabut, Delphine; Bros, Andree; **Fischer, Anne-Marie**
CORPORATE SOURCE: INSERM U428, Universite Paris V, Paris, Fr.
SOURCE: Thrombosis Research (2002), 106(4-5), 213-221
CODEN: THBRAA; ISSN: 0049-3848
PUBLISHER: Elsevier Science Inc.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Fucoidans are **sulfated polysaccharides** extracted from brown marine algae. A purified fucoidan fraction exhibits the same venous **antithrombotic** activity as heparin in rabbits, but with a lower **anticoagulant** effect. Because of its heparin-like structure, we postulated that fucoidan might modulate heparin-binding angiogenic growth factor activity. We thus studied its effect, at **antithrombotic** concns., on fibroblast growth factor (FGF)-2-induced proliferation and differentiation of human umbilical vein endothelial cells. The fucoidan effect on endothelial cell differentiation was evaluated by studying the expression of surface proteins (i.e. integrin, adhesion mol.) known to be modulated by FGF-2 and involved in angiogenesis, and by quantifying closed areas delimited by vascular tubes formed on reconstituted basement membrane. Fucoidan had no modulatory effect on the mitogenic activity of FGF-2, but significantly increased tubular structure d. induced by FGF-2. Fucoidan alone increased α6 integrin subunit expression with only partially organized tubular structure. In the presence of FGF-2, fucoidan enhanced α6, β1 and PECAM-1 and inhibited αvβ3 integrin expression. Heparin had no effect in these systems. The most striking effect of fucoidan was observed on α6 expression and tube formation was abolished by monoclonal anti-α6 antibodies. Fucoidan plus FGF-2 effect on α6 expression was markedly decreased by monoclonal anti-FGF-2 antibodies, indicating that fucoidan acts mainly via FGF-2. These results show that, at **antithrombotic** concns., contrary to heparin, fucoidan can enhance vascular tube formation induced by FGF-2 with a modulation of the expression of surface proteins (mainly α6) involved in angiogenesis.

REFERENCE COUNT: 32 THERE ARE 32 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L23 ANSWER 24 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2001:794800 CAPLUS
DOCUMENT NUMBER: 136:144930
TITLE: Characterization, chemical modifications and in vitro **anticoagulant** properties of an exopolysaccharide produced by Alteromonas infernus
AUTHOR(S): Colliec Jouault, Sylvia; Chevolot, Lionel; Helley, Dominique; Ratiskol, Jacqueline; Bros, Andree; Sinquin, Corinne; Roger, Olivier; **Fischer, Anne-Marie**
CORPORATE SOURCE: Laboratoire de Biochimie et Molecules Marines, Departement Valorisation des Produits, URM2, IFREMER/CNRS (UMR7540, CNRS/Universite Paris 13), Nantes, 44311, Fr.
SOURCE: Biochimica et Biophysica Acta (2001), 1528(2-3), 141-151
CODEN: BBACAQ; ISSN: 0006-3002
PUBLISHER: Elsevier Science B.V.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB A new low-mol.-weight 'heparin-like' component was obtained from an exopolysaccharide produced by a mesophilic strain found in deep-sea hydrothermal vents. Data concerning the structure of the native high-mol.-weight exopolysaccharide (106 g/mol, 10% **sulfate** content) are reported for the first time. Two depolymn. processes were used to obtain low-mol.-weight (24-35+103 g/mol) oversulfated fractions (**sulfate** content 20 or 40%). NMR studies indicated that after **sulfation** (40%), the low-mol.-weight fraction obtained by free radical depolymn. was less **sulfated** in the 6-O-position than the fraction depolymd. by acid hydrolysis. The free radical depolymd. product also had **sulfated** residues in the 4-O-position and disulfated ones in the 2,3-O-positions. Moreover, the compds. generated by the free radical process were more homogeneous with respect to mol. mass. Also for the first time, the **anticoagulant** activity of the low-mol.-weight exopolysaccharide fractions is reported. When the fractions obtained after **sulfation** and depolymn. were compared with heparins, **anticoagulant** activity was detected in oversulfated fractions, but not in native exopolysaccharide. The free radical depolymd. fraction inhibited thrombin generation in both contact-activated and

thromboplastin-activated plasma, showing a prolonged lag phase only in the contact-activated assay. Affinity co-electrophoresis studies suggested that a single population of **polysaccharide** chains binds to **antithrombin** and that only a subpopulation strongly interacts with heparin cofactor II.

REFERENCE COUNT: 46 THERE ARE 46 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L23 ANSWER 25 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2001:167772 CAPLUS
DOCUMENT NUMBER: 134:198049
TITLE: Use of a low molecular weight **sulfated polysaccharides** to obtain a medicine with **antithrombotic** activity
INVENTOR(S): Colliec-Jouault, Sylvia; **Durand, Patrick; Fischer, Anne-Marie; Jozefonvicz, Jacqueline; Letourneur, Didier; Millet, Jean**
PATENT ASSIGNEE(S): Institut Francais de Recherche Pour l'Exploitation de la Mer (IFREMER), Fr.; Centre National de la Recherche Scientifique (CNRS); Universite Rene Descartes (Paris V)
SOURCE: PCT Int. Appl., 33 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: French
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2001015654	A2	20010308	WO 2000-FR2421	20000901
WO 2001015654	A3	20010607		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM			
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
FR 2797768	A1	20010302	FR 1999-10965	19990901
FR 2797768	B1	20030613		
EP 1207891	A2	20020529	EP 2000-960777	20000901
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL				

PRIORITY APPLN. INFO.: FR 1999-10965 A 19990901
WO 2000-FR2421 W 20000901

AB The invention concerns the use of a **sulfated polysaccharide** capable of being obtained by radical depolymn. of a raw **fucan** derived from Pheophycea, said **polysaccharide** having a molar mass not more than 10.000 g/mol, to obtain a medicine for preventing or treating vascular **thrombosis**, in particular venous **thrombosis**, arterial **thrombosis** and arterial restenosis. **Fucan** from Ascophyllum nodosum (mol. weight >600,000) was depolymd. with copper acetate, diafiltered, concentrated, lyophilized, and reduced to obtain a **sulfated polysaccharide** having mol. weight <500. The antithroombgotic activity of this **polysaccharide** was shown in rabbits and rats.

L23 ANSWER 26 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 2001:167154 CAPLUS
DOCUMENT NUMBER: 134:320526
TITLE: Relationship between **antithrombotic** activities of **fucans** and their structure
AUTHOR(S): Boisson-Vidal, Catherine; Chaubet, Frederic; Chevolot, Lionel; Sinquin, Corinne; Theveniaux, Jocelyne; **Millet, Jean**; Sternberg, Claude; Mulloy, Barbara; **Fischer, Anne Marie**
CORPORATE SOURCE: Unite de Recherche Marine 2 CNRS/IFREMER, Laboratoire de Recherches sur les Macromolecules (UMR 7540), Universite Paris-Nord, Villetaneuse, Fr.
SOURCE: Drug Development Research (2000), 51(4), 216-224
CODEN: DDREDK; ISSN: 0272-4391
PUBLISHER: Wiley-Liss, Inc.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB A low mol. weight **fucan** fraction extracted from the brown seaweed Ascophyllum nodosum was previously shown to exhibit dose-related venous **antithrombotic** activity with an ED80 of about 20 mg/kg, 2 h after a single s.c. injection HCII. Its activity was comparable to that of a low mol. weight heparin (Dalteparin). This **fucan** fraction is one of several, with a range of different structure parameters, prepared by degradation of the whole native **fucan**. These low mol. weight fractions were compared using a Wessler stasis **thrombosis** model in rabbits and by determination of their in vitro and ex vivo **anticoagulant** activities. I.v. administrations of these fractions reduced **thrombosis** in a dose-dependent manner. Partial removal of **sulfate** groups and/or partial degradation lead to a significant decrease in their **anticoagulant** and **antithrombotic** activities. The integrity of the regular pattern of **sulfation** of the fucoidan is necessary for **antithrombotic** activity.

REFERENCE COUNT: 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L23 ANSWER 27 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 2000:627453 CAPLUS

DOCUMENT NUMBER: 133:294208

TITLE: Modulation of vascular human endothelial and rat smooth muscle cell growth by a fucosylated chondroitin **sulfate** from echinoderm

AUTHOR(S): Tapon-Bretaudiere, J.; Drouet, B.; Matou, S.; Mourao, P. A. S.; Bros, A.; **Letourneur, D.**; Fischer, A. M.

CORPORATE SOURCE: Laboratoire d'Hematologie, CHU Necker, INSERM U428, Universite Paris V, Paris, 75743, Fr.

SOURCE: Thrombosis and Haemostasis (2000), 84(2), 332-337
CODEN: THHADQ; ISSN: 0340-6245

PUBLISHER: F. K. Schattauer Verlagsgesellschaft mbH

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Fucosylated chondroitin **sulfate** is a glycosaminoglycan extracted from the sea cucumber Ludwigothurea grisea. This **polysaccharide** has the same structure as a mammalian chondroitin **sulfate** but some of the glucuronic acid residues display **sulfated** fucose branches. Anti-coagulant and **antithrombotic** properties of fucosylated chondroitin **sulfate** have already been described. In order to further investigate its potential therapeutic use as an **antithrombotic** agent, we studied its effect on vascular smooth muscle cell (SMC) proliferation and endothelial cell proliferation, migration and Tissue Factor Pathway Inhibitor (TFPI) release. The expts. were performed on SMC from rat thoracic aorta and on human umbilical vein endothelial cell (HUVEC) in culture with or without added fibroblast growth factors (FGF-1 and FGF-2). Our results showed that: (i) fucosylated chondroitin **sulfate** had a strong inhibitory effect on SMC proliferation (IC50 = 10 ± 5 μg/mL) and (ii) no effect on HUVEC proliferation and migration assays, in the absence of exogenous FGF, while heparin had inhibitory effects; (iii) fucosylated chondroitin **sulfate** (10 μg/mL) enhanced FGF-1 and FGF-2 induced HUVEC proliferation by 45% (145.4 ± 7.2%) and 27% (126.9 ± 4.2%), resp.; (iv) on FGF-induced HUVEC migration, fucosylated chondroitin **sulfate** (10 μg/mL) had a strong enhancing effect with FGF-1, +122% (222.2 ± 15.8%), three times higher than that of heparin, and a lower enhancing effect with FGF-2, +43% (142.7 ± 4.6%), whereas heparin had no effect; (v) fucosylated chondroitin **sulfate** stimulated TFPI release, mainly on the free form, +98% (198.2 ± 25.%). In addition, the structural features of the **polysaccharide** associated with its biol. activity were resolved using chemical modified fucosylated chondroitin **sulfates**. **Sulfated** fucose branches groups are essential to the potentiating effect of the **polysaccharide** on HUVEC proliferation and migration. Surprisingly, removal of fucose branches from the fucosylated chondroitin **sulfate** did not abolish TFPI release. Finally, partial reduction of the glucuronic acid carboxyl groups limited the potentiating effect on HUVEC proliferation and migration but did not affect TFPI release. In conclusion, this fucosylated chondroitin **sulfate** from invertebrate origin reveals useful properties for an **antithrombotic** agent: inhibition of SMC proliferation, enhancement of endothelium wound repair and TFPI release. These properties on vascular cells, associated with a low bleeding tendency and an **antithrombotic** activity, strongly suggest its potential use as a new therapeutic agent in arterial **thrombosis** and restenosis, with a more favorable effect than heparin.

REFERENCE COUNT: 47 THERE ARE 47 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L23 ANSWER 28 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1999:191646 CAPLUS
DOCUMENT NUMBER: 130:217984
TITLE: **Antithrombotic** and **anticoagulant** activities of a low molecular weight fucoidan by the subcutaneous route
AUTHOR(S): **Millet, Jean**; **Colliec, S.**; Mauray, S.; Theveniaux, J.; Sternberg, C.; Boisson Vidal, C.; Fischer, A. M.
CORPORATE SOURCE: Laboratories Fournier, Dijon, Fr.
SOURCE: Thrombosis and Haemostasis (1999), 81(3), 391-395
CODEN: THHADQ; ISSN: 0340-6245
PUBLISHER: F. K. Schattauer Verlagsgesellschaft mbH
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Fucoidans (high-mol.-weight **sulfated polysaccharides** extracted from brown seaweeds) have **anticoagulant** and **antithrombotic** effects. They inhibit thrombin by catalyzing both serpins (**antithrombin** and heparin cofactor II) according to their chemical structures and origins. A low-mol.-weight (LMW) fucoidan of 8 kDa was obtained by chemical degradation of a high-mol.-weight fraction. The **antithrombotic** and **anticoagulant** activities of this new compound were compared to those of a low-mol.-weight heparin (LMWH), dalteparin, following s.c. administration to rabbits. This LMW fucoidan exhibited dose-related venous anti-thrombotic activity, with an ED80 of 20 mg/kg, 2 h after a single s.c. injection. Its activity was comparable to that of dalteparin (200 anti-Xa IU/kg) and was maximal 30 min after a single s.c. injection. The activity remained stable (70%) 1-4 h after injection, but disappeared by 8 h. The anti-thrombotic activity was not associated with either a prolongation of the thrombin clotting time (TCT) or an increase in anti-Xa activity, contrary to dalteparin. A slight prolongation of APTT occurred with both compds. This venous **antithrombotic** activity was associated with a decrease in ex viva thrombin generation and with a significant increase in the lag phase in a thrombin generation test. LMW fucoidan thus has potent **antithrombotic** activity and a potentially weaker hemorrhagic effect (i.e. a smaller effect on coagulation tests and a smaller prolongation of the bleeding time) than dalteparin.
REFERENCE COUNT: 28 THERE ARE 28 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L23 ANSWER 29 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1999:35343 CAPLUS
DOCUMENT NUMBER: 130:108198
TITLE: Modulation of human endothelial cell proliferation and migration by fucoidan and heparin
AUTHOR(S): Giraux, Jean-Luc; Matou, Sabine; Bros, Andree; Tapon-Bretaudiere, Jacqueline; **Letourneur, Didier**; **Fischer, Anne-Marie**
CORPORATE SOURCE: Lab. Hematologie, Hospital Necker-Enfants Malades, Paris, F-75743, Fr.
SOURCE: European Journal of Cell Biology (1998), 77(4), 352-359
CODEN: EJCBDN; ISSN: 0171-9335
PUBLISHER: Gustav Fischer Verlag
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Fucoidan is a **sulfated polysaccharide** extracted from brown seaweeds. It has **anticoagulant** and **antithrombotic** properties and inhibits, as well as heparin, vascular smooth muscle cell growth. The authors investigated, in the presence of serum and human recombinant growth factors, the effects of fucoidan and heparin on the growth and migration of human umbilical vein endothelial cells (HUVEC) in culture. The authors found that fucoidan stimulated fetal bovine serum-induced HUVEC proliferation, whereas heparin inhibited it. In the presence of fibroblast growth factor-1 (FGF-1), both fucoidan and heparin potentiated HUVEC growth. In contrast, fucoidan and heparin inhibited HUVEC proliferation induced by FGF-2, hut did not influence the mitogenic activity of vascular endothelial growth factor (VEGF). In the in vitro migration assay from a denuded area of confluent cells, the 2 **sulfated polysaccharides** markedly enhanced the migration of endothelial cells in the presence of FGF-1. A weak inhibitory effect on cell migration was found only with the 2 **polysaccharides** at high concns. (≥100 μg/mL) in presence of serum or combined with FGF-2. The results indicated that heparin and fucoidan can be used as tools to further investigate the cellular mechanisms regulating the proliferation and migration of human vascular cells. The data already suggest a potential role of fucoidan as a new therapeutic agent of vegetal

origin in the vascular endothelium wound repair.
REFERENCE COUNT: 39 THERE ARE 39 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L23 ANSWER 30 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1998:662228 CAPLUS
DOCUMENT NUMBER: 129:270387
TITLE: Fucoidan, as heparin, induces tissue factor pathway inhibitor release from cultured human endothelial cells
AUTHOR(S): Giraux, Jean-Luc; Tapon-Bretaudiere, Jaqueline; Matou, Sabine; **Fischer, Anne-Marie**
CORPORATE SOURCE: Lab. Hematologie, Hopital Necker-Enfants Malades, Paris, F-75743, Fr.
SOURCE: Thrombosis and Haemostasis (1998), 80(4), 692-695
CODEN: THHADQ; ISSN: 0340-6245
PUBLISHER: F. K. Schattauer Verlagsgesellschaft mbH
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Fucoidan, a **sulfated polysaccharide** extracted from brown seaweeds, has **antithrombotic** properties, the mechanism of which is not yet completely understood. The authors demonstrated that fucoidan, as heparin, induces tissue factor pathway inhibitor (TFPI) release from cultured human umbilical vein endothelial cells (HUVEC). The TFPI accumulation in the HUVEC supernatants depends on the incubation time and **polysaccharide** concentration After 30-60 min of incubation, TFPI concentration (total antigen level) was twice higher in the presence of both **polysaccharides** than in their absence. After 1 h of incubation, in the presence of increasing concns. of each **polysaccharide**, an optimal stimulation was observed for 0.5 μg/mL of fucoidan and 5 μg/mL of heparin, as evidenced by a raise of the basal TFPI level: a 2-fold increase for the total antigen and a 3-fold increase for the free antigen. These data suggest that TFPI released from vascular endothelial cells may contribute to the **antithrombotic** effect of fucoidan.

L23 ANSWER 31 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1998:579330 CAPLUS
DOCUMENT NUMBER: 130:1626
TITLE: Mechanism of factor IXa inhibition by **antithrombin** in the presence of unfractionated and low molecular weight heparins and fucoidan
AUTHOR(S): Mauray, Sandrine; de Raucourt, Emmanuelle; Talbot, Jean-Claude; Dachary-Prigent, Jeanne; Jozefowicz, Marcel; **Fischer, Anne-Marie**
CORPORATE SOURCE: Hopital Necker Enfants-Malades, Laboratoire de Recherche en Hematologie, Universite Paris V, Paris, 75743, Fr.
SOURCE: Biochimica et Biophysica Acta (1998), 1387(1-2), 184-194
CODEN: BBACAQ; ISSN: 0006-3002
PUBLISHER: Elsevier Science B.V.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Heparin exerts its **anticoagulant** activity by catalyzing the inhibition of coagulation proteases by **antithrombin** (AT). Its main target is thrombin but it also catalyzes the inhibition of the other serine-proteases of the coagulation cascade, such as factor IXa (fIXa). The aim of this study was to compare the catalysis of inhibition of blood fIXa by **antithrombin** in the presence of several **sulfated polysaccharides** with **anticoagulant** activity, i.e. heparin, three low mol. weight heparins (LMWH) widely used in therapeutics, and fucoidan. Plots of the second-order rate consts. of the fIXa-**antithrombin** reaction vs. the concentration of added heparin and LMWH are bell-shaped and fit the kinetic model established for thrombin-**antithrombin** reaction by R. Jordan, et al. (1979, J. Biol. Chemical, 254, 2902-2913). In the ascending branch, the catalyst (C) binds quickly to the inhibitor (I) to form a catalyst-inhibitor (CI) complex which is more reactive toward the enzyme (E) than the free inhibitor, leading to the formation of an inactive enzyme-inhibitor complex (EI) and the release of free catalyst, in a rate-limiting second step. After a maximum corresponding to an optimal catalyst concentration, the decrease in the reaction rate was in keeping with the formation of a catalyst-enzyme (CE) complex, whose inactivation by the CI complex was slower than that of the free enzyme. Maximum second-order rate consts. for the inhibition of fIXa by AT were 105, 6.8, 12.24 and 22 μM-1 min-1 with heparin, Enoxaparin, Fraxiparin and Fragmin, resp., leading to 3500-, 225-, 405- and 728-fold increases in the inhibition rate in the absence of **polysaccharide** , resp. Fucoidan yielded 23-fold increase in the fIXa-

antithrombin interaction rate. The kinetic profiles obtained with this **polysaccharide** exhibited ascending branch which correlated well with the kinetic model based on the formation of binary complexes (CI or CE). Fucoidan was covalently conjugated with a fluorescent probe (DTAF) and used in conjunction with fluorescence anisotropy to follow its binding to **antithrombin**, heparin cofactor II (HCII), thrombin and fIXa. The binding of fucoidan to these proteins occurred with low affinities when compared to heparin and LMWH. Fucoidan had higher affinity for the inhibitor HCII compared to **antithrombin** and enzymes. These data suggest that binding of heparins and fucoidan to the inhibitor (CI) is required for the **polysaccharide**-dependent enhancement in the rate of neutralization of the enzyme by the inhibitor.

REFERENCE COUNT: 38 THERE ARE 38 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L23 ANSWER 32 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1995:673194 CAPLUS
DOCUMENT NUMBER: 123:257145
TITLE: Thioxyloside derivatives as orally active venous **antithrombotics**
AUTHOR(S): Bellamy, Francois; Barberousse, Veronique; Martin, Niall; Masson, Philippe; **Millet, Jean**; Samreth, Soth; Sepulchre, Christiane; Theveniaux, Jocelyne; Horton, Derek
CORPORATE SOURCE: Laboratories Fournier, Daix, 21121, Fr.
SOURCE: European Journal of Medicinal Chemistry (1995), 30(Suppl., Proceedings of the 13th International Symposium on Medicinal Chemistry, 1994), 101s-15s
CODEN: EJMCA5; ISSN: 0223-5234
PUBLISHER: Elsevier
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The synthesis and pharmacol. evaluation of several thioxylosides, especially naroparcil, beciparcil and iliparcil, are discussed. These thioxylosides are remarkably good substrates of glycosyltransferase I and in vivo, after oral administration, they elicit a large increase in the plasma-level concentration of glycosaminoglycans (GAGs). At least 20% of the circulating GAG is dermatan **sulfate**-like material capable of inhibiting thrombin via HC-II. It was also demonstrated that a large proportion of these GAGs are built on the thioxyloside substrate. Finally, the thioxylosides had potent **antithrombotic** activity in rats.

L23 ANSWER 33 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1995:366566 CAPLUS
DOCUMENT NUMBER: 122:151093
TITLE: The effect of the β-D-xyloside naroparcil on circulating plasma glycosaminoglycans. An explanation for its known **antithrombotic** activity in the rabbit
AUTHOR(S): Masson, Philippe J.; Coup, Dominique; **Millet, Jean**; Brown, Neil L.
CORPORATE SOURCE: Centre de Recherche et Developpement, Laboratoires Fournier S.C.A., Daix, 21121, Fr.
SOURCE: Journal of Biological Chemistry (1995), 270(6), 2662-8
CODEN: JBCHA3; ISSN: 0021-9258
PUBLISHER: American Society for Biochemistry and Molecular Biology
DOCUMENT TYPE: Journal
LANGUAGE: English

AB β-D-Xylosides are known to initiate or prime free glycosaminoglycan (GAG) chain synthesis in cell and tissue culture. As such, the effect of the venous **antithrombotic** β-D-xyloside, naroparcil, was investigated on the plasma GAG profile in the rabbit after oral administration. Using dose-response expts., the authors showed that **antithrombin** activity via **antithrombin** III and heparin cofactor II was increased in parallel with GAG plasma levels compared to control. A more detailed qual. examination of plasma GAGs by cellulose acetate electrophoresis and ion-exchange chromatog., following oral administration of naroparcil at 400 mg/kg, revealed the presence of higher d. charged mols. compared to control. The extracted GAGs were found to activate inhibition of thrombin by heparin cofactor II and contained approx. 25% of a dermatan **sulfate**-like compound (undetectable in control), which could be responsible for the **antithrombotic** effect. Using radiolabeled naroparcil, the authors found radiolabeled GAG fractions and the fact that naroparcil was a substrate for galactosyltransferase I, the second enzyme responsible for GAG chain polymerization, suggested that the compound could initiate in vivo the biosynthesis of **antithrombotic** free GAG chains. This is, to the authors knowledge, the first description of

the in vivo effect of a β-D-xyloside on GAG biosynthesis; furthermore, this is correlated with an **antithrombotic** action.

L23 ANSWER 34 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1995:288295 CAPLUS
DOCUMENT NUMBER: 122:71666
TITLE: The venous **antithrombotic** profile of naroparcil in the rabbit
AUTHOR(S): **Millet, Jean**; Theveniaux, Jocelyne; Brown, Neil L.
CORPORATE SOURCE: Fournier S.C.A. Laboratories, Daix, Fr.
SOURCE: Thrombosis and Haemostasis (1994), 72(6), 874-9
CODEN: THHADQ; ISSN: 0340-6245
PUBLISHER: Schattauer
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The venous **antithrombotic** profile of naroparcil or (4-[4-cyanobenzoyl]-phenyl)-1,5-dithio-β-D-xylopyranoside was investigated in the rabbit following single i.v. and oral administration. Naroparcil attenuated thrombus development in a Wessler stasis model of venous **thrombosis** (jugular vein) employing bovine factor Xa as a thrombogenic stimulus giving ED50 values of 21.9 mg/kg and 36.0 mg/kg after resp. i.v. and oral administration. Venous **antithrombotic** activity was maximal 2-3 h after i.v. administration and 4-8 h after oral administration. Four hours after the oral administration of maximal **antithrombotic** (Wessler model, factor Xa) doses (100 and 400 mg/kg), naroparcil had no significant effect on bleeding time. In platelet poor plasma obtained from animals treated 4 h previously with various doses (25 to 400 mg/kg) of naroparcil, there was no detectable antifactor Xa nor **antithrombin** activity. Similarly, naroparcil had no effect on APTT nor on thrombin time. A sensitized thrombin time (to about 35 s) was modestly but significantly increased following oral administration of the compound at 400 mg/kg. However, thrombin generation by the intrinsic pathway was reduced in a dose-related manner, maximal reduction being 655 at 400 mg/kg. The same doses of naroparcil enhanced the formation of thrombin/heparin cofactor II complexes at the expense of thrombin/**antithrombin** III complexes in plasma incubated with (125I)-human-α-thrombin and induced the appearance of dermatan **sulfate**-like material in the plasma of treated rabbits, as measured by a heparin cofactor II-mediated thrombin inhibition assay. The results suggest that naroparcil could have a safe venous **antithrombotic** profile following oral administration (**antithrombotic** effect compared to bleeding risk). It is probably that part of the mechanism of action of the β-D-xyloside, naroparcil, is due to the induction of chondroitin **sulfate**-like glycosaminoglycan biosynthesis, this material being detectable in the plasma.

L23 ANSWER 35 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1992:51169 CAPLUS
DOCUMENT NUMBER: 116:51169
TITLE: **Anticoagulant** properties of a fucoidan fraction
AUTHOR(S): **Colliec, S.**; Fischer, A. M.; Tapon-Bretaudiere, J.; Boisson, C.; Durand, P.; Jozefonvicz, J.
CORPORATE SOURCE: LRM, Univ. Paris Nord, Villetaneuse, 93430, Fr.
SOURCE: Thrombosis Research (1991), 64(2), 143-54
CODEN: THBRAA; ISSN: 0049-3848
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Fucoidans are high-mol.-weight (8 + 105-106) **sulfated polysaccharides** widely dispersed in brown seaweed cell walls. When extracted from several brown algae, they show **anticoagulant** properties. The chemical degradation of a crude extract from Pelvetia canaliculata was used to obtain a low-mol.-weight **polysaccharide** (.apprx.20,000) for possible clin. use. Its **anticoagulant** potency was investigated through the inhibition of factor IIa and factor Xa in the presence of **antithrombin** III or heparin cofactor II. The degraded fucoidan revealed a potent **antithrombin** activity. Studied in an **antithrombin** III-depleted plasma or in the presence of purified heparin cofactor II, the fucoidan was as efficient as heparin and dermatan **sulfate** on heparin cofactor II potentiation, at the same concentration by weight In whole plasma or in the presence of the purified inhibitor, an anti-factor IIa activity mediated by **antithrombin** III was 30-fold less potent than that of heparin on a weight basis. No antifactor Xa activity was detected in the presence of the degraded fucoidan.

L23 ANSWER 36 OF 36 CAPLUS COPYRIGHT 2004 ACS on STN
ACCESSION NUMBER: 1991:124842 CAPLUS
DOCUMENT NUMBER: 114:124842
TITLE: **Sulfated polysaccharides, anticoagulant** and anticomplementary agent prepared from **fucans** from brown seaweeds and process for obtaining them
INVENTOR(S): **Colliec, Sylvia**; Bretaudiere, Jacqueline; Durand, Patrick; Fischer, Anne Marie; Jozefonvicz, Jacqueline; Kloareg, Bernard; Vidal, Catherine
PATENT ASSIGNEE(S): Institut Francais de Recherche pour l'Exploitation de la Mer (IFREMER), Fr.
SOURCE: Eur. Pat. Appl., 17 pp.
CODEN: EPXXDW
DOCUMENT TYPE: Patent
LANGUAGE: French
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 403377	A1	19901219	EP 1990-401636	19900613
EP 403377	B1	19951206		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE				
FR 2648463	A1	19901221	FR 1989-7857	19890614
FR 2648463	B1	19930122		
WO 9015823	A1	19901227	WO 1990-FR420	19900613
W: AU, CA, JP, KP, KR, SU, US				
AU 9058410	A1	19910108	AU 1990-58410	19900613
CN 1051564	A	19910522	CN 1990-104927	19900613
DD 296937	A5	19911219	DD 1990-341619	19900613
JP 04506089	T2	19921022	JP 1990-508929	19900613
JP 3042543	B2	20000515		
EP 676207	A2	19951011	EP 1995-105774	19900613
EP 676207	A3	19951108		
EP 676207	B1	20000906		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE				
AT 131176	E	19951215	AT 1990-401636	19900613
AT 196089	E	20000915	AT 1995-105774	19900613
ES 2152998	T3	20010216	ES 1995-105774	19900613
US 5321133	A	19940614	US 1992-778220	19920116

PRIORITY APPLN. INFO.:
FR 1989-7857 A 19890614
EP 1990-401636 A3 19900613
WO 1990-FR420 A 19900613

AB The title **polysaccharides**, with mol. weight 5000-40,000 and containing <0.15% protein contaminants, are prepared from **fucans** extracted from Pheophyces and contain more S than the original **fucans**. **Fucans** extracted from shoots of brown seaweeds were heated at concentration 10 mg/mL in 1N H_2SO_4 at 45° until **fucan** hydrolysis was complete, concentrated, subjected to ultrafiltration and lyophilized. The lyophilizate (500 mg in 5 mL 0.2M NaCl) was subjected to gel chromatog. and fractions were combined and subjected to ultrafiltration to give fractions with mol. weight 1300-700,000, S content 7.2-11%, and protein content <0.01-0.85%, which had **anticoagulant** activity 1.3-6.6 IU/mg.